

A NEW FIXATIVE FOR CUTANEOUS HISTOPATHOLOGY

(FORMALIN-NICKEL)*

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The fact that the skin consists of several dissimilar layers offers several technical problems to the histopathologist. One of the principal difficulties concerns the selection of a fixative that penetrates quickly and preserves equally well all the different cellular and tissular elements from the stratum corneum of the epidermis to the adipose tissue of the hypoderm. It is essential to choose a good and practical fixative that fulfills the named requirements and also can be washed out easily after fixation is completed.

In our histopathological laboratory we use, as a fixative for every day routine work as well as for scientific investigations, an aqueous solution of 8% neutral formalin (neutralized with magnesium or calcium carbonate) with 1% nickel nitrate ($\text{Ni}(\text{NO}_3)_2 + 6\text{H}_2\text{O}$; Nickelous Nitrate, Purified, Crystals; Merck).

Formula of "Formalin-Nickel" fixative for one liter of the solution:

Nickel nitrate.....	10 gr
Neutral formalin.....	80 cc
Distilled water.....	920 cc

Because this fixative is absolutely stable, one or several liters are prepared at one time. For a small skin biopsy specimen we use about 15 to 30 cc of the solution (that is 50 times the biopsy volume). At least 24 hours are needed for routine fixation. But the specimens can remain in the fixative for two or more days and even, without any problem, for months because no precipitates will appear. The tissue can be used afterwards for embedding or frozen sections. After the fixation the specimen is washed in running water during 3-4 hours; and one or more hours in distilled water, changed several times. It may be embedded in paraffin, celloidin, etc. For the paraffin method the tissue is dehydrated by passing it through a series of rising alcohols (50%; 70%; 80%; 90%; 96% and 100%) alcohol-xytol, (or benzol or toluol); pure xytol (or benzol, etc.).

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It is then embedded in paraffin according to the classic method.

The sections, after being deparaffinized, may be studied with a great variety of stains for investigation of any component of the epidermis, corium or hypoderm. We consider the "formalin-nickel" fixative as the ideal fixative for the study and investigation of the elastic fibers (Weigert; Taenzer-Unna; Gomori; etc.). Also collagen and pre-collagen stain excellently with their appropriate and respective technics (Van Gieson; Mallory; Unna; etc.). It is also a good fixative for the study of the nervous system (Bielschowsky-Gross etc.). All the cellular elements of all the epidermic layers (stratum germinativum, stratum spinosum, etc.) such as the tonofibrils, the cytoplasm, the nucleus, the mitochondria, Golgi apparatus, can be studied.

Shape, stainability, and structure of the different tissue and cellular elements of the skin retain their normal aspect and can be studied in detail with the proposed fixative.

This fixative is being used in the laboratory of General Pathology of the University for all organs, with results that confirm our investigations (personal communication of Professor Oliva Otero).

SUMMARY

Aqueous solution of 8% neutralized formalin with 1% nickel nitrate is described and recommended as a fixative ("Formalin-Nickel") in cutaneous histopathology. It is a good fixative for routine work, since it is handled easily and rapidly: can be preserved indefinitely, and produces good staining of the epidermis and the dermis (H-E; Mallory, Giemsa Van Gieson; Mayer's hemalum; Regaud's and Heidenhain's iron-hematoxylin; etc.). It is an excellent fixative of elastic fibers, assuring investigation of these fibers with excellent staining (Weigert, Gomori, Taenzer-Unna, etc.) and detail. It is a certain fixative of the neurovegetative system of the skin. It allows one to keep the biopsy specimens for many years without changing their capacity for silver staining (Bielschowsky-Gross, Bielschowsky-Jabonero, etc.).